CHRONIC TOXICITY SUMMARY

ETHYLBENZENE

(Phenylethane; NCI-C56393)

CAS Registry Number: 100-41-4

I. Chronic Toxicity Summary

Inhalation reference exposure level	**2000 µg/m^3 (400 ppb)**
Critical effect(s)	Liver, kidney, pituitary gland in mice and rats
Hazard index target(s)	Alimentary system (liver); kidney; endocrine system

II. Physical and Chemical Properties (HSDB, 1994)

Description	colorless liquid
Molecular formula	C_8H_{10}
Molecular weight	106.16 g/mol
Boiling point	136.2°C
Melting point	-95°C
Vapor pressure	10 torr @ 25.9°C
Density	0.867 g/cm^3 @ 20°C
Solubility	Soluble in ethanol and ether, low solubility in water (0.014 g/100 ml at 15°C)
Conversion factor	1 ppm = 4.35 mg/m^3

III. Major Uses or Sources

Ethylbenzene is used as a precursor in the manufacture of styrene (HSDB, 1994). It is also used in the production of synthetic rubber, and is present in automobile and aviation fuels. It is found in commercial xylene (Reprotext, 1994). In 1996, the latest year tabulated, the statewide mean outdoor monitored concentration of ethylbenzene was approximately 0.4 ppb (CARB, 1999a). The latest annual statewide emissions from facilities reporting under the Air Toxics Hot Spots Act in California, based on the most recent inventory, were estimated to be 161,846 pounds of ethylbenzene (CARB, 1999b).

IV. Effects of Human Exposure

Studies on the effects of workplace exposures to ethylbenzene have been complicated by concurrent exposures to other chemicals, such as xylenes (Angerer and Wulf, 1985). Bardodej

and Cirek (1988) reported no significant hematological or liver function changes in 200 ethylbenzene production workers over a 20-year period.

V. Effects of Animal Exposure

Rats and mice (10/sex/group) were exposed to 0, 100, 250, 500, 750, and 1000 ppm (0, 434, 1086, 2171, 3257, and 4343 mg/m^3) ethylbenzene 6 hours/day, 5 days/week for 90 days (NTP, 1988; 1989; 1990). Rats displayed significantly lower serum alkaline phosphatase in groups exposed to 500 ppm or higher. Dose-dependent increases in liver weights were observed in male rats beginning at 250 ppm, while this effect was not seen until 500 ppm in the females. An increase in relative kidney weights was seen in the 3 highest concentrations in both sexes. Minimal lung inflammation was observed in several of the treatment groups, but this phenomenon was attributed to the presence of an infectious agent rather than to ethylbenzene exposure. The mice in this study did not show any treatment-related effects except for elevated liver and kidney weights at 750 and 1000 ppm, respectively.

Rats and mice were exposed to ethylbenzene (greater than 99% pure) by inhalation for 2 years (NTP, 1999; Chan *et al*., 1998). Groups of 50 male and 50 female F344/N rats were exposed to 0, 75, 250, or 750 ppm, 6 hours per day, 5 days per week, for 104 weeks. Survival of male rats in the 750 ppm group was significantly less than that of the chamber controls. Mean body weights of 250 and 750 ppm males were generally less than those of the chamber controls beginning at week 20. Mean body weights of exposed groups of females were generally less than those of chamber controls during the second year of the study. In addition to renal tumors, the incidence of renal tubule hyperplasia in 750 ppm males was significantly greater than that in the chamber controls. The severity of nephropathy in 750 ppm male rats was significantly increased relative to the chamber controls. Some increases in incidence and severity of nephropathy were noted in all exposed female rats, but these were statistically significant only at 750 ppm.

Groups of 50 male and 50 female B6C3F1 mice were exposed to 0, 75, 250, or 750 ppm ethylbenzene by inhalation, 6 hours per day, 5 days per week, for 103 weeks. Survival of exposed mice was similar to controls. Mean body weights of females exposed to 75 ppm were greater than those of the chamber controls from week 72 until the end of the study. In addition to lung and liver tumors, the incidence of eosinophilic liver foci in 750 ppm females was significantly increased compared to that in the chamber controls. There was a spectrum of nonneoplastic liver changes related to ethylbenzene exposure in male mice, including syncytial alteration of hepatocytes, hepatocellular hypertrophy, and hepatocyte necrosis. The incidences of hyperplasia of the pituitary gland pars distalis in 250 and 750 ppm females and the incidences of thyroid gland follicular cell hyperplasia in 750 ppm males and females were significantly increased compared to those in the chamber control groups. Based on an evaluation of all the non-cancer data in mice and rats OEHHA staff selected 75 ppm as the NOAEL for the NTP (1999) study.

Rats (17-20 per group) were exposed to 0, 600, 1200, or 2400 mg/m^3 for 24 hours/day on days 7 to 15 of gestation (Ungvary and Tatrai, 1985). Developmental malformations in the form of "anomalies of the uropoietic apparatus" were observed at the 2400 mg/m^3 concentration.

Skeletal retardation was observed in all exposed groups compared with controls. The incidence of skeletal abnormalities increased with higher concentrations of ethylbenzene.

Rabbits exposed by these investigators to the same concentrations as the rats on days 7 to 15 of gestation, exhibited maternal weight loss with exposure to 1000 mg/m^3 ethylbenzene. There were no live fetuses in this group for which abnormalities could be evaluated. No developmental defects were observed in the lower exposure groups.

Rats (78-107 per group) and rabbits (29-30 per group) were exposed for 6 or 7 hours/day, 7 days/week, during days 1-l9 and 1-24 of gestation, respectively, to 0, 100, or 1000 ppm (0, 434, or 4342 mg/m^3) ethylbenzene (Andrew *et al.*, 1981; Hardin *et al.*, 1981). No effects were observed in the rabbits for maternal toxicity during exposure or at time of necropsy. Similarly, no effects were seen in the fetuses of the rabbits. The only significant effect of ethylbenzene exposure in the rabbits was a reduced number of live kits in the 1000 ppm group. A greater number and severity of effects were seen in rats exposed to 1000 ppm ethylbenzene. Maternal rats exposed to 1000 ppm exhibited significantly increased liver, kidney, and spleen weights compared with controls. Fetal rats showed an increase in skeletal variations at the 1000 ppm concentration, but the results of the 100 ppm exposure were not conclusive.

Clark (1983) found no significant effects on body weight, food intake, hematology, urinalysis, organ weights or histopathology in rats (18 per group) exposed to 100 ppm (434 mg/m^3) ethylbenzene for 6 hours/day, 5 days/week, for 12 weeks.

Degeneration of the testicular epithelium was noted in guinea pigs and a rhesus monkey exposed to 600 ppm (2604 mg/m^3) for 6 months (Wolf *et al.*, 1956). No effects were reported for female monkeys exposed to the same conditions.

Cragg *et al.* (1989) exposed mice and rats (5/sex/group) to 0, 99, 382, and 782 ppm (0, 430, 1659, and 3396 mg/m^3) 6 hours/day, 5 days/week for 4 weeks. Some evidence of increased salivation and lacrimation was seen in the rats exposed to 382 ppm. No other gross signs of toxicity were observed. Both male and female rats had significantly enlarged livers following exposure to 782 ppm. Female mice also showed a significant increase in liver weight at this concentration. No histopathological lesions were seen in the livers of these mice.

Dose-dependent induction of liver cytochrome P450 enzymes in rats by ethylbenzene was observed by Elovaara *et al.* (1985). Rats (5 per group) were exposed to 0, 50, 300, or 600 ppm (0, 217, 1302, or 2604 mg/m^3) ethylbenzene for 6 hours/day, 5 days/week for 2, 5, 9, or 16 weeks. Cytochrome P450 enzyme induction, and microscopic changes in endoplasmic reticulum and cellular ultrastructure were evident at all ethylbenzene concentrations by week 2, and persisted throughout the exposure. Liver weights were not elevated in these studies.

VI. Derivation of the Chronic Reference Exposure Level

Study	NTP, 1999; Chan *et al*., 1998
Study population	Male and female rats and mice (50 per group)
Exposure method	Discontinuous inhalation
Critical effects	Nephrotoxicity, body weight reduction (rats) hyperplasia of the pituitary gland; liver cellular alterations and necrosis (mice)
LOAEL	250 ppm
NOAEL	75 ppm
Exposure continuity	6 hours/day, 5 days/week
Exposure duration	103 weeks.
Average experimental exposure	13 ppm for NOAEL group
Human equivalent concentration	13 ppm for NOAEL group (gas with systemic effects, based on RGDR = 1.0 using default assumption that lambda (a) = lambda (h))
LOAEL uncertainty factor	1
Subchronic uncertainty factor	1
Interspecies uncertainty factor	3
Intraspecies uncertainty factor	10
Cumulative uncertainty factor	30
Inhalation reference exposure level	0.4 ppm (400 ppb; 2 mg/m^3; 2,000 $\mu g/m^3$)

The REL is based on a lifetime toxicity/carcinogenesis study. The NOAEL for non-neoplastic effects in the study was 75 ppm, and the LOAEL was 250 ppm. Some shorter duration studies discussed above (e.g. NTP, 1988, 1989, 1990) identify higher concentrations as NOAELs, but the study used (NTP 1999) is the most recent available and is considered the most reliable for assessing chronic effects.

U.S. EPA based its RfC on developmental toxicity studies in rats and rabbits (Andrew *et al.*, 1981; Hardin *et al.*, 1981; U.S. EPA, 1994). The NOAEL in the studies was 100 ppm, and the LOAEL was 1000 ppm. In accordance with its methodology, U.S. EPA did not use a time-weighted average concentration for the discontinuous exposure experiment since the key effect was developmental toxicity. If OEHHA methodology is followed (which includes the time-weighted averaging of the exposure concentrations, and uncertainty factors of 3 (interspecies, with RGDR = 1) and 10 (intraspecies), this study would indicate a REL of 0.6 ppm (3 mg/m^3). The study by Ungvary and Tatrai (1985) reported a NOAEL of 600 mg/m^3 for developmental and maternal effects in several species. However, the reporting and general quality of this paper create less confidence in its results.

For comparison to the proposed REL of 0.4 ppm, Clark (1983) found no significant effects in rats exposed to 100 ppm ethylbenzene 6 h/day, 5 d/week, for 12 weeks. This NOAEL can be time-adjusted to 18 ppm, then divided by a subchronic UF of 3, an interspecies UF of 3, and an intraspecies UF of 10 which results in a REL of 0.2 ppm. (The default value of 1 for RGDR was used). It appears that the proposed REL provides a sufficient margin of safety to provide

protection against the reported developmental effects (Andrew *et al.,* 1981; Hardin *et al.,* 1981; Ungvary and Tatrai, 1985)

VII. Data Strengths and Limitations for Development of the REL

The strengths of the inhalation REL for ethylbenzene include the availability of controlled exposure inhalation studies in multiple species at multiple exposure concentrations and with adequate histopathogical analysis, and the observation of a NOAEL in lifetime chronic inhalation exposure studies. The major area of uncertainty is the lack of adequate human exposure data.

VIII. References

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